

AMENDMENTS TO THE CLAIMS

1. (Currently amended) A method for quantitation of zinc ion in a sample comprising:

i) disposing in said sample a donor fluorescent sulfonamide fluorophore and a carbonic anhydrase protein having conjugated thereto an acceptor fluorophore, wherein the binding of said donor fluorophore to the conjugated carbonic anhydrase is dependent upon the presence of zinc ion complexed to the conjugated carbonic anhydrase;

ii) measuring the intensity of a ~~photoluminescence~~-fluorescence emission of the acceptor fluorophore upon excitation of the donor fluorophore at a first excitation wavelength;

iii) measuring the intensity of a ~~photoluminescence~~-fluorescence emission of the acceptor fluorophore upon excitation of the acceptor fluorophore at a second excitation wavelength;

iv) obtaining the ratio of the intensities measured in steps ii) and iii) and;

v) relating the quantity of zinc ion in the sample to the ratio obtained in step iv).

2. (Canceled)

3. (Canceled)

4. (Original) The method of claim 2, in which the donor fluorophore is Dapoxyl sulfonamide.

5. (Currently amended) The method of ~~claim 2~~, claim 1, in which the acceptor fluorophore is ALEXA-FLUOR 594 pyrano[3,2-g:5,6-g']diquinolin-13-ium, 6-[2-carboxyl-4 (or 2-carboxyl-5) [carboxamido-N-pentyl-N'-maleimide]phenyl]-1,2,10,11-tetrahydro-1,2,2,10,10,11-hexamethyl-4,8-bis(sulfomethyl); inner salt.

6. (Canceled)

7. (Canceled)

8. (Currently amended) The method of ~~claim 2~~, claim 1, in which the carbonic anhydrase is human carbonic anhydrase II having a cysteine residue substituted for ~~leucine residue 198~~ serine residue 166 or having a cysteine residue substituted for histidine 36 and the acceptor fluorophore is conjugated to said cysteine residue.

9. (Canceled)

10. (Canceled)

11. (Currently amended) The method of claim 5, in which the carbonic anhydrase is human carbonic anhydrase II having a cysteine residue substituted for ~~leucine residue 198~~ serine

residue 166 or having a cysteine residue substituted for histidine 36 and the acceptor fluorophore is conjugated to said cysteine residue.

12. (Canceled)

13. (Canceled)

14. (Currently amended) A method for quantitation of zinc ion in a sample comprising:

i) disposing in said sample a donor fluorescent sulfonamide fluorophore and a fusion protein comprising a carbonic anhydrase protein and a fluorescent protein acceptor fluorophore, wherein the binding of said donor fluorophore to the fusion protein is dependent upon the presence of zinc ion complexed to the ~~conjugated~~-carbonic anhydrase domain of the fusion protein;

ii) measuring the intensity of a ~~photoluminescence~~-fluorescent emission of the acceptor fluorophore upon excitation of the donor fluorophore at a first excitation wavelength;

iii) measuring the intensity of a ~~photoluminescence~~-fluorescent emission of the acceptor fluorophore upon excitation of the acceptor fluorophore at a second excitation wavelength;

iv) obtaining the ratio of the intensities measured in steps ii) and iii) and;

v) relating the quantity of zinc ion in the sample to the ratio obtained in step iv).

15. (Canceled)

16. (Original) The method of claim 15, in which the fluorescent protein is Green Fluorescent Protein, Enhanced Green Fluorescent Protein, Yellow Fluorescent Protein, DsRed, or Blue Fluorescent Protein.

17. (Canceled)

18. (Canceled)

19. (Original) The method of claim 16, in which the donor fluorophore is Dapoxyl sulfonamide.

20. (Original) The method of claim 14, in which the sample comprises a living cell.

21. (New) The method of claim 1, in which the donor fluorophore is selected from the group consisting of dansylamide and ABDN.

22. (New) The method of claim 1, in which the acceptor fluorophore is rhodamine maleimide.

23. (New) The method of claim 21, in which the acceptor fluorophore is rhodamine maleimide.

24. (New) The method of claim 14, in which the donor fluorophore is selected from the group consisting of dansylamide and ABDN.